Cytologia **48**: 795–801, 1983

Cytological Studies in the Genus *Tephrosia* Pers.

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Received January 8, 1982

Genus *Tephrosia* belongs to tribe Galegeae of family Fabaceae. It is represented by 400 species (Hutchinson 1964) which are distributed in warmer regions of the world especially Africa and tropical Australia. Very little cytological work has been done so far (about 12%) in the genus and previous studies mainly involved chromosome counts. In view of this an attempt has been made to study meiosis and root tip mitosis in available species of this genus.

Material and methods

Material of eight species was obtained in the form of seed from different sources which are listed in Table 1. Plants were raised in pots at Meerut University Experimental Farm. Dormancy of seed was broken by treating the seed with concentrated sulphuric acid. Voucher specimens are deposited in the Department of Ag. Botany, Meerut University, Meerut.

For meiosis, flower buds were collected in forenoon, fixed for at least 24 hours in Carnoy's fluid (absolute alcohol: chloroform: acetic acid, 6: 3: 1) and stored in 70% ethyl alcohol. Anthers were squashed in 2.0% acetocarmine.

For mitosis, young and healthy root tips were pretreated with saturated solution of α-bromonaphthalene for about one and a half to two hours, fixed in acetic alcohol (1: 3) for 24 hours and squashed in 2.0% acetocarmine.

Photomicrographs were made from temporary preparations and measurements (length and width of chromosomes) were made with the help of Olympus micrometer eye piece. TC1% and TF% were calculated as

$$\text{TC1}\% = \frac{\text{Total length of a chromosome pair}}{\text{Total length of chromosome complement}} \times 100$$

$$\text{TF}\% = \frac{\text{Total length of all short arms}}{\text{Total length of chromosome complement}} \times 100$$

Results

1. *Meiosis*

Meiosis was studied in eight species (Figs. 1–10). Chromosome associations and chiasmata frequencies are presented in Table 1. It is obvious that all the collections were diploids having $n=11$ (Table 1).

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Table 1. Gametic chromosome numbers and chiasmata frequencies of 8 species of *Tephrosia* Pers. along with their sources

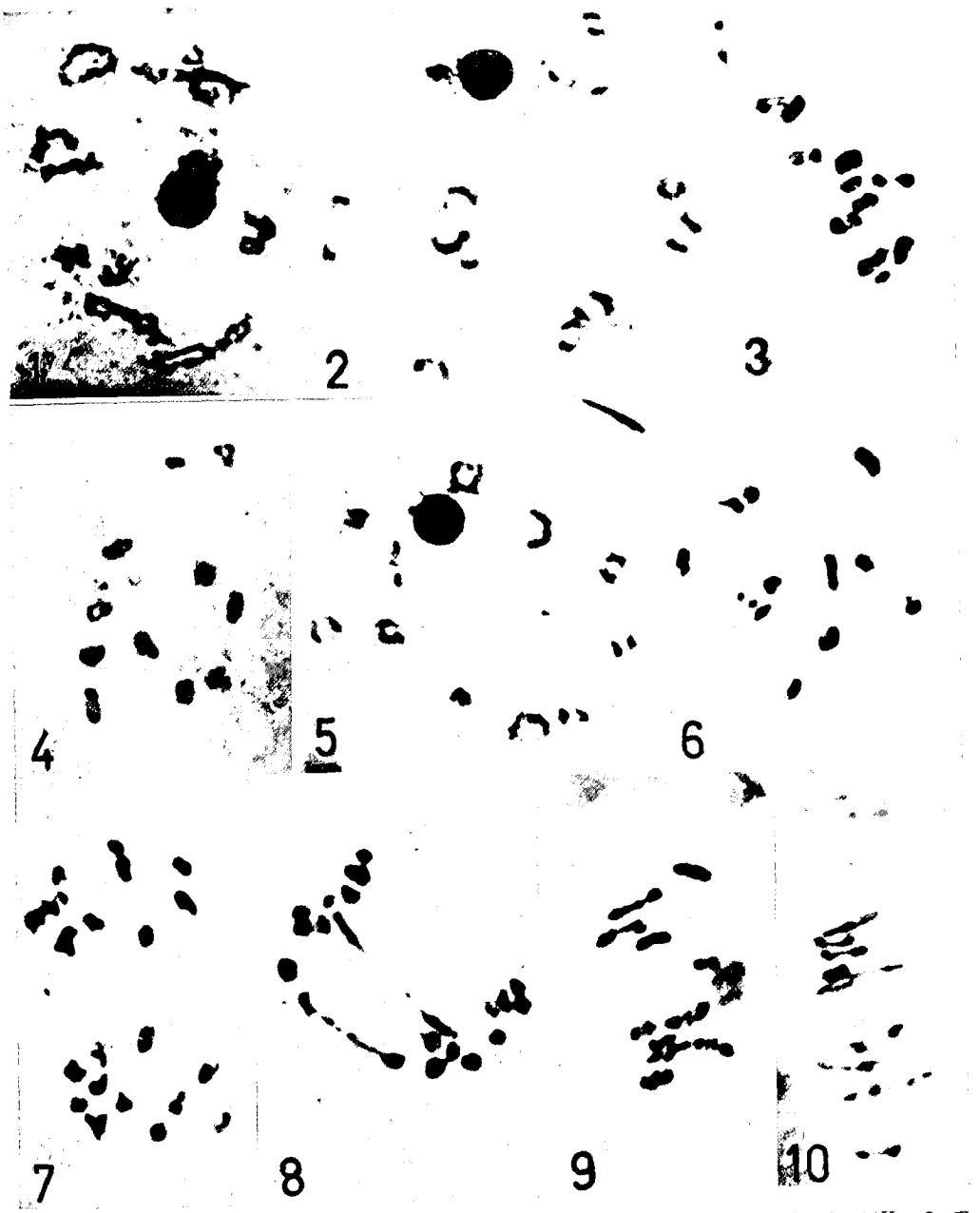
Species	Coll. no.	Gametic number (*n*)	Chiasmata		Source	Accession number given by source
			Xta/PMC	Xta/bivalent		
T. adunca Berth	1822	11	—	—	Southern Regional Station, Georgia, U. S. A.	308580
T. bracteolata Guill. et Perr.	1823	11	16.25	1.47	CSIRO, Canberra, Australia	CPI 36719
T. maxima Pers.	1824	11	21.00	1.99	CSIRO, Canberra, Australia	CPI 36384
T. noctiflora Bojev ex Baker	1825	11	19.65	1.79	CSIRO, Canberra, Australia	CPI 24092
T. oxygona Welw. ex Baker	1826	11	20.30	1.84	Southern Regional Station, Georgia, U. S. A.	185583
T. polystachya E. Mey	1827	11	19.83	1.80	Southern Regional Station, Georgia, U. S. A.	365043
T. purpurea L. Pers.	1828	11	17.62	1.60	CSIRO, Canberra, Australia	CPI 36720
T. villosa Pers. var. *incana*	1829	11	20.58	1.87	Southern Regional Station, Georgia, U. S. A.	

Table 2. Analysis of mitotic chromosomes in three species of *Tephrosia* (in each species, the first row represents absolute lengths of chromosome; the second row gives arm ratios; the third row gives relative chromosome lengths and the fourth row gives TCI%)

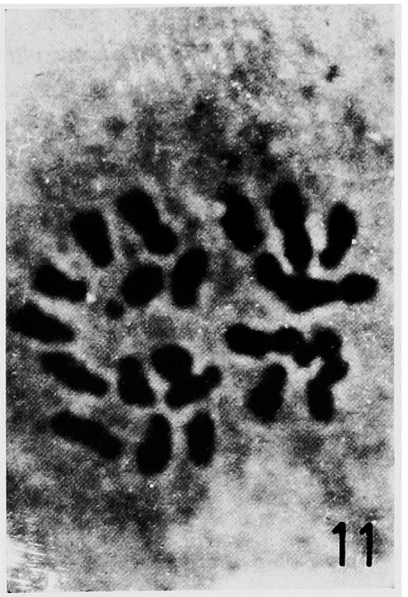
Species	Chromosome pairs										
	1	2	3	4	5	6	7	8	9	10	11
T. bracteolata	3.04	2.97	2.97	2.48	2.48	2.35	2.21	2.07	1.93	1.93	1.72
	1.20	1.39	1.26	2.00	1.57	2.40	2.20	2.00	1.80	1.54	1.50
	100.00	97.73	97.73	81.82	81.82	77.27	72.73	68.18	63.64	63.64	56.82
	11.61	11.34	11.34	9.50	9.50	8.97	8.44	7.91	7.39	7.39	6.60
T. noctiflora	2.55	2.35	2.00	1.86	1.72	1.72	1.66	1.59	1.52	1.52	1.45
	1.69	1.83	1.64	1.25	1.27	1.27	1.78	1.87	1.00	1.00	1.62
	100.00	91.89	78.38	72.97	67.56	67.56	64.86	62.16	59.46	59.46	56.76
	12.80	11.76	10.03	9.34	8.65	8.65	8.30	7.96	7.61	7.61	7.22
T. purpurea	2.00	1.93	1.79	1.66	1.38	1.38	1.31	1.10	1.10	1.10	1.03
	1.42	1.15	1.67	2.00	1.50	1.00	1.11	1.00	1.00	1.00	2.00
	100.00	96.55	89.65	82.57	68.96	68.96	65.52	55.17	55.17	55.17	51.72
	12.66	12.23	11.35	10.48	8.73	8.73	8.30	6.99	6.99	6.99	6.55

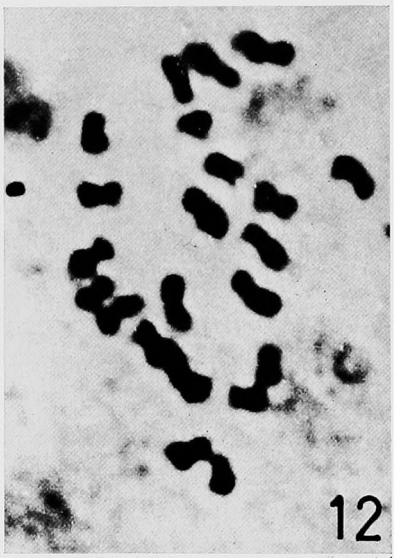
2. *Mitosis and karyotypes*

Three species of *Tephrosia* namely, *T. bracteolata*, *T. noctiflora* and *T. purpurea* were analysed for katyotypes. Eleven pairs of chromosomes were recorded in each species. Photomicrographs of somatic metaphase plates are presented in Figs.



Figs. 1–10. Meiosis in different *Tephrosia* species. 1, *T. adunca*, early diakinesis, 11^{II}. 2, *T. bracteolata*, diakinesis, 11^{II}. 3, *T. maxima*, metaphase I, 11^{II}. 4, *T. noctiflora*, prometaphase I, 11^{II}. 5, *T. oxygona*, diakinesis, 11^{II}. 6, *T. polystachya*, metaphase I, 11^{II}. 7, *T. polystachya*, anaphase I, 11:11. 8, *T. polystachya*, anaphase I, showing late disjunction. 9, *T. purpurea*, metaphase I, 11^{II}. 10. *T. villosa* var. *incana*, metaphase I, 11^{II}.


11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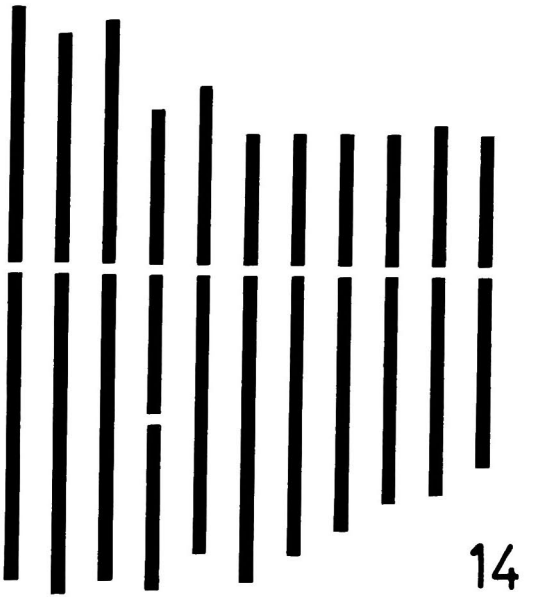
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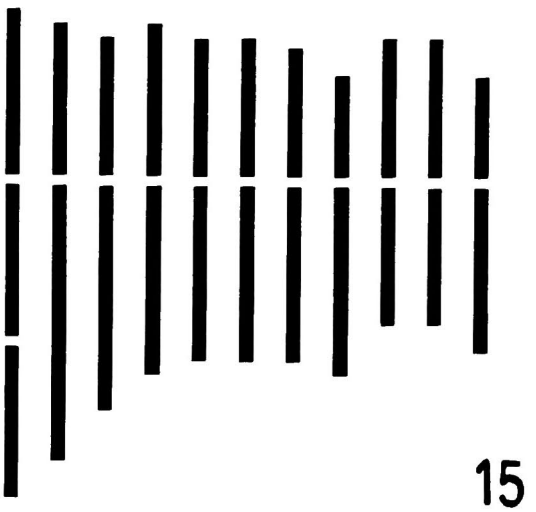
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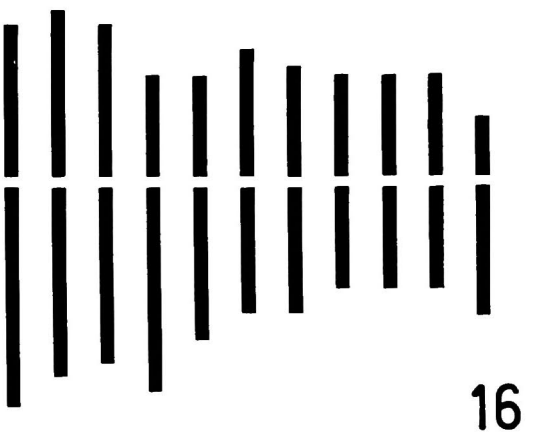
16

11–13 and data on chromosome measurements and chromosome morphology are presented in Tables 2–4. Corresponding idiograms are shown in Figs. 14–16.

Chromosomes were designated as 1–11, according to decreasing lengths. Depending upon their absolute length, chromosomes were classified into three categories, namely A=more than 3.0 μ, B=1.5 μ–3.0 μ and C=less than 1.5 μ.

The chromosomes were further subdivided according to position of centromere and chromosome formulae are presented in Table 4.

Discussion

A survey of previous literature on cytological studies in the genus *Tephrosia* deomonstrated that only 47 of the 400 known species of the genus have been cytologically worked out so far of which 46 species exhibit $2n=22$. In the present study also

Table 3. Chromosome data in three species of *Tephrosia*

Species	Total chromatin length (μ)	Mean chromosome length (μ)	Longest/ shortest ratio	TF%	Chromatin volume (μ^3)
T. bracteolata	26.15	2.38	1.77	37.99	22.02
T. noctiflora	19.94	1.81	1.75	40.83	12.59
T. purpurea	15.78	1.43	1.94	44.10	8.48

Table 4. Karyotypic formulae of three species of *Tephrosia* (A, B and C represent long; medium and short chromosome respectively; 'sc' used as subscript represents secondary constriction in long arm; superscript 'm', 'sm' and 'st' represent respectively the median, submedian and subterminal position of centromeres)

S. no.	Species	Karyotypic formulae
1.	*T. bracteolata*	$1A^{sm}+1scB^{sm}+7B^{sm}+2B^{st}$
2.	*T. noctiflora*	$1scB^{sm}+6B^{sm}+2C^{m}+2C^{sm}$
3.	*T. purpurea*	$1B^{m}+3B^{sm}+5C^{m}+2C^{sm}$

all the eight species examined exhibited $2n=22$. However, scattered chromosome counts of $2n=32$ (Kawakami 1930), $2n=24$ (Ramanathan 1950, 1955, Bhatt 1976) and $2n=44$ (Tandon and Malik 1960) are also known in the genus. In the tribe Galegeae to which *Tephrosia* belongs, the predominant base unmber is $x=8$ and $x=11$ is mainly found in the genera *Astragalus*, *Psoralea*, *Mundulea* and *Tephrosia*. Atchison (1951) has suggested that from tribe Galegeae either the genera with $x=10$, 11 should be transferred to Dalbergieae or else these two tribes should be merged. Such a view was later supported by Turner and Fearing in 1959. Burkart (1952) suggested on morphological grounds that some genera from Galegeae having predominantly woody members may be transferred to Dalbergieae. In view of this,

Figs. 11–16. 11–13: Mitotic metaphase plates in different *Tephrosia* species. 11, *T. bracteolata*. 12, *T. noctiflora*. 13, *T. purpurea*. 14–16: Idiograms prepared from mitotic metaphase in different *Tephrosia* species. 14, *T. bracteolata*. 15, *T. noctiflora*. 16, *T. purpurea*.

since *Tephrosia* has $x=11$, according to Atchison's (1951) recommendation, it should be transferred from Galegeae to Dalbergieae. But since the members of the genus *Tephrosia* are mainly herbs and shrubs, according to recommendations of Burkart (1952), it should be retained in Galegeae. It can, therefore, be concluded that we should not reassign genera to different tribes on purely morohological and purely cytological grounds.

It is, however, important to notice that *Tephrosia* is the only genus, where $2n=22$ is almost the exclusive chromosome number and, therefore, has unique position in this respect. Evolutionary significance of this situation is not very clearly known so far except that the members of this genus may be related to members of tribe Dalbergieae more closely than to members of Galegeae.

A survey of cytological literature indicated that no studies on karyotypes in this genus were earlier undertaken. The present study of karyotypes in three species indicated that average chromosome length varied, which was 2.37 μ in *T. bracteolata*, 1.81 μ in *T. noctiflora* and 1.43 μ in *T. purpurea*. When classified with respect to karyotype asymmetry (Stebbins 1971), *T. noctiflora* was placed in 1A and the remaining two species were placed in 2A indicating that perhaps *T. bracteolata* and *T. purpurea* are more advanced relative to *T. noctiflora*.

Tephrosia has been divided by Hooker (1879) into three subgenera—*Macronyx*, *Brissonia* and *Reineria*. While *Macronyx* includes annuals with simple leaves, *Brissonia* includes shrubs and *Reineria* includes perennial herbs. If perennial habit is more primitive as some morphologists believe, *T. purpurea* which belongs to *Reineria*, should then be a primitive species. Karyotype asymmetry though worked out only in three species during the present study, suggests that *T. purpurea* may be more advanced.

Acknowledgements

The authors are grateful to various agencies listed in Table 1 for the supply of seed material which made it possible for us to conduct this study.

Summary

1. In the genus *Tephrosia* male meiosis was studied in 8 species and mitosis with the help of root tip was studied in three species. The chromosome number was invariably found to be $n=11$ and $2n=22$. Chromosome number in three species (*T. maxima* Pers., *T. oxygona* Welw. ex Baker and *T. polystachya* E. Mey) were reported for the first time.
2. The meiosis was normal in all the cases. The chiasmata frequency/PMC varied from 16.15 (*T. bracteolata*) to 21.00 (*T. maxima*).
3. The total chromatin length varied from 15.78 μ (*T. purpurea*) to 26.15 μ (*T. bracteolata*).
4. The chromosomes were mainly metacentric and did not vary greatly so that the level of asymmetry was low.

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